

**EXTRACTION OF FEMALE CRUDE SEX PHEROMONE OF ONION
ARMYWORM *Spodoptera exigua* HÜBNER (LEPIDOPTERA:
NOCTUIDAE) AND ITS ATTRACTANCY EFFECT**

RYAN ALEXIS P. BAUTISTA

An Undergraduate Thesis Submitted to the faculty of the Department Crop
Protection College of Agriculture, Central Luzon State University
Science City of Munoz, Nueva Ecija, Philippines
in Partial Fulfilment of the requirement
for the Degree of

**BACHELOR OF SCIENCE IN AGRICULTURE
(Crop Protection)**

SEPTEMBER 2019

ACCEPTANCE SHEET

This undergraduate manuscript entitled "EXTRACTION OF FEMALE CRUDE SEX PHEROMONE OF ONION ARMYWORM *Spodoptera exigua* HÜBNER (LEPIDOPTERA: NOCTUIDAE) AND ITS ATTRACTANCY EFFECT," prepared and submitted by RYAN ALEXIS P. BAUTISTA, in partial fulfillment of the requirements for the degree of BACHELOR OF SCIENCE IN AGRICULTURE (CROP PROTECTION), is hereby accepted:


MARILYN G. PATRICIO, Ph.D.
Adviser


ELAIDA R. FIE. GALAN, Ph.D.
Critic

Date Signed

Sept 2019

Date Signed


CELYNNE O. PADILLA, M.Sc.
Department Research Coordinator

Date Signed

Accepted as partial fulfillment of the requirements for the degree of BACHELOR OF SCIENCE IN AGRICULTURE (CROP PROTECTION):


RONALDO T. ALBERTO, Ph.D.
Department Chairperson


MARIA LUISA T. MASON, Ph.D.
College Research Coordinator

Date Signed

Date Signed


ERNESTO A. MARTIN, Ph.D.
Dean, College of Agriculture

Date Signed

BIOGRAPHICAL SKETCH

The author is the first child of four children of Reysie Bautista-Okamoto. He was born on January 27, 1996 at Mandaluyong General Hospital. He finished his kindergarten at Our Lady of the Sacred Heart Academy (OLSHA) at Guimba, Nueva Ecija in the year 2002. He studied at the same school for elementary education and finished in year 2008. He finished his high school education at Our Lady of the Sacred Heart College (OLSHCO) at year 2012. He took up Bachelor of Science in Animal Husbandry in Central Luzon State University (CLSU) Science City of Munoz Nueva Ecija but he shifts to another course Bachelor of Science in Agriculture Major in Crop Protection with field of specialization in Entomology.

ACKNOWLEDGEMENT

This study would not have been accomplished without the help of every individual behind as they serve as an inspiration and help to the author. Therefore, the author would like to express his deepest and sincere thanks and appreciation to those who shared their time and efforts towards the success of this study

Above all and anyone else, to the one who guided his paths and gave light in every direction that he took and for all the courage that he gave when the author is down to rise up again. For all the helping hands, the Almighty Father gave when the author almost gave up. To all the people that helped, the author, this would not be possible without His grace and love.

This study would not be possible without the support, knowledge, patient and understanding of Dr. Marilyn G. Patricio. The author is sincerely thankful for being his thesis adviser.

To Mr. Jose Emmanuel de Luna, who stood as thesis adviser in the retirement of Dr. Patricio; the author is thankful for the advice and time allotted to finish the study.

To Dr. Bonifacio F. Cayayab for his kindness, guidance and advices for the success of the study.

To the project team of Dr. Navasero of National Crop Protection Council for giving some reading material for the enhancement of the study.

To Dr. Sally Rafael for giving the author some knowledge about the study and solvents used in extraction.

To his critic, Dr. Elaida R. Fiegalan for her kindness, guidance and criticism for the enhancement of the paper.

To the then Department chairperson Dr. Marita S. Labe for giving the motivation for facing bigger challenges in life; and the incumbent chairperson, Dr. Ronaldo T. Alberto for assistance in ensuring the final defense will proceed and for guiding the author in finishing the manuscript

To the Department of Crop Protection Faculty and Former Faculty for giving inspirational messages, creative suggestions, help and support that the author considers creative criticism for a better version. To the staff, Ms. Sheryl Marcha for giving supportive message and providing the materials during the conduct or study.

To his Out-Reach Philippines Family, to his former boss Tori O. Robertson and Felina S. Delfin. To all OPI staff Tita Ami, Tita Ems, Ate Krizza, Ate Rose, Kuya Elvis and Kuya Rhed for giving the author chance to be a part your family.

To his Gamma Sigma Scorpion Family especially to his batch mates, brothers, and sisters for giving unconditional laughter and joy. The author is grateful to find companions and second family in his journey in CLSU.

To his Pest Management Society Family for helping and believing on each other.

To his Entomates for their endless support and endless laughters that led to everlasting friendship.

To kuya Miguelito Isip for letting the author join the surveillance of armyworm.

To Philscat Family specially to Ate Mamay, Sir Val, Ate Leila and Mary Grace for supporting and giving criticism to be better

Most of all the author is sincerely thankful to the owners of the onion farmers that let him used their field for collection and trapping armyworm, Manong Fred Policarpio, Nanay Filipina Salavation, Mr. Manny dela Cruz, Mr. Danilo Cucal, and Ate Melody Torres.

TABLE OF CONTENTS

	PAGE
LIST OF TABLES	ix
LIST OF FIGURE	x
LIST OF APPENDICES	xi
ABSTRACT	xiii
INTRODUCTION	
Importance of the Study	2
Statement of the Problem	3
Objectives of the Study	4
Scope and Limitations of the Study	4
Time and Place of the study	5
REVIEW OF RELATED LITERATURE	
Distribution of <i>Spodoptera exigua</i>	6
Migration of <i>Spodoptera exigua</i>	6
Biology of <i>Spodoptera exigua</i>	
Egg	7
Larva	7
Pupa	7
Adult	8
Damage of <i>Spodoptera exigua</i>	8
Host of <i>Spodoptera exigua</i>	9
Kinds of Pheromones	10
Sex Pheromone as Monitoring Tool for IPM	13
Advantage and Disadvantage of Sex Pheromone Traps	14
Extraction of Crude Sex Pheromones	14
Methodology of Sex Pheromone Application	15
METHODOLOGY	
Collection of Onion Armyworm	17
Mass Rearing of Onion Armyworm	17
Extraction of Crude Pheromone	20

Evaluation of Extracted Crude Sex Pheromone	
Laboratory Condition	22
Field Condition	24
Data Gathered	
Under Laboratory Condition	28
Under Field Condition	28
Statistical Analysis	29
RESULTS AND DISCUSSION	
Laboratory Condition	
Percentage Attractancy	31
Field Condition	
Brgy. Dolores Sto. Domingo, Nueva Ecija	35
Brgy. San Fabian Sto. Domingo, Nueva Ecija	35
Brgy. San Francisco Sto. Domingo Nueva Ecija	36
Number of Armyworm Larva and Damage Rating	40
SUMMARY, CONCLUSIONS, AND RECOMMENDATIONS	
Summary	42
Conclusions	42
Recommendations	42
LITERATURE CITED	43
APPENDICES	46

LIST OF TABLES

TABLE		PAGE
1	Crop susceptible to armyworm	9
2	Different solvent used in extracting crude sex pheromone of onion armyworm	20
3	Geographic location in three Brgys. of Sto. Domingo, Nueva Ecija used for trapping site for male armyworm moth	24
4	Damage rating used for armyworm	28
5	Average percent attractancy of male moth using different solvent in no choice method	32
6	Number of trapped male moth onion armyworm moth in onion field at Barangay Dolores Sto. Domingo, Nueva Ecija using crude pheromone extract and live virgin female moth	37
7	Number of trapped male moth onion armyworm moth in onion field at Barangay San Fabian Sto. Domingo, Nueva Ecija using crude pheromone extract and live virgin female moth	38
8	Number of trapped male moth onion armyworm moth in onion field at Barangay San Francisco Sto. Domingo, Nueva Ecija using crude pheromone extract and live virgin female moth	39
9	Number of larva and damage rating of armyworm monitored in three Brgys. of Sto. Domingo, Nueva Ecija monitored during the trapping	41

LIST OF FIGURES

FIGURE		PAGE
1	Rearing process of onion armyworm: a. collection of armywormegg and larva in the field, b. rearing of egg and larva on the container, c. mating of adult moth on hollow tube, d. newly hatched eggs.	18
2	Separation of male and female <i>Spodoptera exigua</i> : a. male pupa, b. female pupa, c. male moth, d. female moth	19
3	Extraction of crude sex pheromone: a. immobilized female armyworm, b. removal of female armyworm 2 abdominal segments containing the pheromone gland, c. Extracted crude sex pheromone	21
4	Diagram of the improvised cylinder used for laboratory test: a. entrance of the cylinder, b. bait, c. rubber septum	23
5	Improvised bait for no choice method	23
7	Point of trap placement in the onion field located at a. Dolores b. San Fabian c. San Francisco,Sto. Domingo, Nueva Ecija	25
8	Set-up of pheromone traps using: a. crude sex pheromone using treatment hexane 1:1 ratio and b. live virgin female moth	26
9	Trapping sites of armyworm in Sto. Domingo, Nueva Ecija: Barangay Dolores, b. Barangay San Fabian, C. Barangay San Francisco	27

LIST OF APPENDICES

APPENDIX		PAGE
1.1a	Square root transformed data of percent attractancy of male moth at 5 min	48
1.1b	Analysis of variance for square root transformed of percent attractancy of male moth at 5 min	48
1.2a	Square root transformed data of percent attractancy of male moth at 10 min	49
1.2b	Analysis of variance for square root transformed data of percent attractancy of male moth at 10 min	49
1.3a	Square root transformed data of percent attractancy of male moth at 15 min	50
1.3b	Analysis of variance for square root transformed data of percent attractancy of male moth at 15 min	50
1.4a	Square root transformed data of percent attractancy of male moth at 20 min	51
1.4b	Analysis of variance for square root transformed data of percent attractancy of male moth at 20 min	51
1.5a	Square root transformed data of percent attractancy of male moth at 25 min	52
1.5b	Analysis of variance for square root transformed data of percent attractancy of male moth at 20 min	52
1.6a	Square root transformed data of percent attractancy of male moth at 30 min	53
1.6b	Analysis of variance for square root transformed data of percent attractancy of male moth at 30 min	53
1.7a	Pooled mean of percent attractancy of male moth	54
1.7b	Analysis of variance for pooled mean of percent attractancy of male moth	54

1.8	Agrometeorological data in Sto. Domingo, Nueva Ecija during trapping of male moth	55
1.9	Number of larva and damage rating of sample plants monitored in three Brgys. of Sto. Domingo, Nueva Ecija monitored during the trapping	56

ABSTRACT

BAUTISTA, RYAN ALEXIS P., Department of Crop Protection, College of Agriculture, Central Luzon State University, Science City of Muñoz, Nueva Ecija, Philippines. September 2019. **EXTRACTION OF FEMALE CRUDE SEX PHEROMONE OF ONION ARMYWORM *Spodoptera exigua* HÜBNER (LEPIDOPTERA: NOCTUIDAE) AND ITS ATTRACTANCY EFFECT**

Adviser: Marilyn G. Patricio, Ph.D.

The study was conducted to evaluate different solvents in extracting crude sex pheromones in female onion armyworm to determine the attractancy effect of the crude sex pheromone to male armyworm moths under laboratory condition and to test the efficacy of the extracted crude pheromones hormones under field condition. The protocol for extraction was based on Cayabyab (1997). The eighth and ninth segments of the abdomen of the female armyworm consisting the pheromone gland were excised, then soaked in solvent (Acetone, Hexane and Ethanol), then placed with rubber septum and the crude pheromone was extracted. The attractancy test was done using no choice method and 20 active male moths (one to two day old) were used per replication. The percent attracted male moth was recorded at 5 min, 10 min, 15 min, 20 min, 25 min and 30 min after released. Treatment 3 (Hexane 1:1) was found significantly attractive on the male moth under laboratory condition after 30 min of observation time with 30% attractancy and Treatment 6 (Ethanol 1:2) was the least with 11.67% attractancy. Number of male moth caught per trap was monitored daily for one week. Also, number of armyworm larval and damage rating were recorded from 30 sample onion plants

Keywords: sex pheromone, attractancy effect, extraction, onion armyworm

LITERATURE CITED

- AGUDA R. (1977). Effect of Attacus Juvenile Hormone on the Common Cutworm *Spodoptera litura* F. (Doctoral dissertation) University of the Philippines, Los Baños. 8-11.
- ARIDA G.S., B.S. PUNZAL, C.C RAVINA JR, V.P. GAPUD, E. RAJOTTE & E. TALER. (2002). Sex Pheromone in Pest Management Monitoring Adult Density of *Spodoptera litura* and *Spodoptera exigua* in Rice Onion Cropping System. 28(8), 23-29.
- AZIDAH A. A., & M. S. AZIRUN. (2006). Life History of *Spodoptera exigua* (Lepidoptera: Noctuidae) on various host plant. 19(1): 613-618.
- BAKER T.C, L.K. GASTON, M. M. POPE, L. S. KUENEN & R. S. VETTER. (1981). A High-Efficiency Collection Device for Quantifying Sex Pheromone Volatilized From Female Glands and Synthetic Sources. 161-168.
- BAKER T.C., W. FRANCKE, J.G.MILLAR, C. LOFTSEDT, B. HANSSON, J.W. DU, P.L. PHELAN, R.S. VETTER, R. YOUNGMAN, & J.L TODD. (1991). Identification and Bioassay of Sex Pheromone Component of Crab Moth *Ectomyelois ceratoniae* Z. 19(1): 54-60.
- BAKER T.C. (2011). Insect Pheromones: Useful Lessons for Crustacean Pheromone Program. 54 (3), 60- 65.
- BUTIN R.G., LEE D.R., WILSON D.M., & MCPHERSON R.M. (2000). Fall Armyworm Damage Assesment Procedure.
- CANDANO R.N. (2017). Progress Report on Mass Rearing of *Spodoptera exigua* on the Laboratory.
- CAYABYAB B.F. (1997). Monitoring and Computer Simulation System for Corn Borer *Ostrinia furnacalis*. 1-6. (Doctoral dissertation) University of the Philippines, Los Baños. 1-16.
- CAYABYAB B.F., M.D. EBUENGA, P.G. GONZALES, M.V. NAVASERO, M.M. NAVASERO, M.G. PATRICIO, R.N. CANDANO, M.S. GUERRERO, K.P. ARDEZ, G.A. S. BURGONTO, W. DE PANIS & B.A.G. CAYABYAB. (2017). Seminar in Insect Pheromone in IPM. Regional Crop Protection, PhilRice, Science City of Muñoz, Nueva Ecija.
- CAPINERA J.L. (2014). Beet Armyworm, *Spodoptera exigua* (Hübner) (Insecta:Lepidoptera: Noctuidae). 16(2), 5-18.
- EDMUNDS A.J. (2010). Host Marking Pheromone (HMP) in the Mexican Fruitfly *Anastrepha aludens*. (7), 60- 65.

- GREENBERG S.M., T.W. SAPPINGTON, B.C. LEGASPI, T.X. LIU, & M. SETAMOU. (2001). Feeding and Life History of *Spodoptera exigua* (Lepidoptera Noctuidae) on Different Host Plants. 28(7), 1-15.
- GOMEZ K.A., & A.A GOMEZ. (1984). Statistical Procedures for Agricultural Research. 567- 577
- INSECTICIDE RESISTANCE ACTION COMMITTEE (IRAC). (2012). Strategies for Sustainable Control Beet Armyworm *Spodoptera exigua* Hübner. <http://www.irac-online.org/beetarmyworm>.
- JAYARAJ J. (2013). Controlling Fruitfly Menace in Bitter Gourd. <http://www.thehindu.com/ci-tech/science/controlling-fruit-fly-menace-in-bitter-gourd/article4474059/.com>.
- KANADEA E. ,& D. S. BHATKHANDER. (2016). Extraction of Ginger Oil Using Different Methods and Effect of Solvents, Time and Temperature to Maximize Yield
- KRISHNAIAH K., I.C. PASALU, & N.R.G. VARMA. (2001). Potential of Pheromone in Rice Pest Management. 34(1): 12-17 .
- LI J.X. (1987). Influence of Peanut Cultivars on the Development and Feeding Activity on Common Cutworm *Spodoptera litura* F. (Doctoral dissertation) University of the Philippines, Los Baños 1-11.
- LIBERLES S. D., & L. B. BUCK. (2006). A Second Class of Chemosensory Receptors in the Olfactory Epithelium. 15(1), 645-650.
- MUNICIPAL AGRICULTURAL OFFICE (M.A.O). (2017). Data on Onion Armyworm *Spodoptera exigua*.
- MILLER J. (2002). Manual on Properties of Organic Solvents. 6(3), 15-23.
- MOHAMMED A., A. MASHALY, M. F. ALI, & M.S. AL-KHALIFA. (2012). Trail Pheromones in Pest Control, New Perspective in Plant Protection. 122-139.
- NAVASERO M., M.M NAVASERO, B.F. CAYABYAB, R. CANDANO, M. EUBUENGA , A.S.B. GIDEON , N. M BAUTISTA , E.M. AQUINO, & G.G. GASPAS. (2017). Investigation on the 2016 Outbreak of the Onion Armyworm *Spodoptera exigua* Hübner (Lepidoptera:Noctuidae) in Onion Growing Areas in Luzon.
- PLANTWISE. (2016). Armyworm Hostplants. <http://www.plantwise.org/Beetarmyworm>.
- PHILIPPINE STATISTIC AUTHORITY (P.S.A.). (2016). Major Vegetables and Rootcrops Quarterly Bulletin. 10(1), 55

- PROVINCIAL AGRICULTURAL OFFICE (P.A.O). (2016). Data on Onion Armyworm Outbreak in Nueva Ecija.
- REGINER F., and J. Law. (1968). Insect Pheromone. *Journal of Lipid Research*, 9 (1), 541- 551
- REGIONAL CROP PROTECTION CENTER REGION III (RCPC). (2016). Onion Armyworm Outbreak in Nueva Ecija Terminal Report.
- SEYBOLD J. & S. DONALDSON. (2015). Pheromones in Insect Pest Management. 57(1), 1-2.
- TETSU A. (2013). Pheromone Signaling Methods and Protocols. 67(2), 3-13.
- TUMLINSON J.H., & E.R. MITCHELL,P.E. SONNET. (1981). Sex Phermone Components of the Beet Armyworm *Spodoptera exigua*. 67(2), 198-200.
- VERHEGGEN F.J., E.HAUBRUGE, & M.C. MESCHER. (2009). Alarm Pheromones Chemical Signaling in Response to Danger. 67(2), 12-23
- WISEMEN B.R., and N.W WIDSTROM. (1984). Armyworm Damage Rating in Onion at Various Infestation Levels and Plant Development Stages
- WAKAMURA S., & M. TAKAI. (1988). Control of Beet Armyworm in Open Fields with Sex Pheromone.
- WAKAMURA S., & M. TAKAI. (1990). Control of Beet Armyworm *Spodoptera exigua* with Synthetic Sex Pheromone.
- WALTER S. (2005). Manual on Pheromone Reception. 1(2), 1-26.
- ZHENG X. L., X.P. CHONG, X.P. WANG, & C.L. LEI. (2001). A review of Geographic Distribution, Overwintering and Migration in *Spodoptera exigua* Hübner (Lepidoptera; Noctuidae). 15 (1), 23-37.